

# SAMPLE CHIP ANALYZING DEVICE AND METHOD FOR ANALYZING THE SAME

[Detailed description of the invention]

[Field of the invention]

The present invention relates to a sample chip analyzing device and a method for analyzing the sample chip that analyzes gene generating modes of cells and biological tissues and analyzes antigen and antibody reactions.

[Themes to be solved by the invention]

In the abovementioned use, a sample chip has been used, in which various types of sample probes such as polynucleotide and protein peptide probes like a DNA probe, an RNA probe, etc., are dot-arrayed and fixed on a glass sheet slide at high density (several tens through seven ten thousands pieces per square centimeter).

For example, in the work of analyzing gene generating modes, a test sample to be analyzed, which is extracted from cells and biological tissues, adjusted, and marked with a fluorescent substance, is adhered to respective sampling probes on a sample chip. Where the sampling probe and the test sample to be analyzed are complementary to each other, they are coupled to each other. To the contrary, where they are not complementary to each other, they are not coupled.

After the sample to be analyzed, which has not been coupled to the sampling probe, is washed off by a buffer solution, the surface of the sample chip is optically scanned, and fluorescence from the marked fluorescent substances is detected to specify the sample to be analyzed,

by the sampling probe to which the sample to be analyzed is coupled.

When optically scanning the sample chip, light of an appointed beam diameter is irradiated thereto from a light source, and at the same time an objective lens, which receives light from a sample chip, and the sample chip are caused to move relative to each other in order to scan the entirety thereof, whereby the marked fluorescent substances of the hybridized sample to be analyzed are pumped.

In the abovementioned method, since reflected pumping light of the fluorescent substance is received together with the fluorescence from the fluorescent substance, it is necessary to provide an optical filter to distinguish the fluorescence from the pumping light. However, since the reflected pumping light is remarkably intense in comparison with the fluorescence, the fluorescence detection is likely to be influenced by the pumping light, wherein the fluorescence detection accuracy was not satisfactory.

Although it is necessary to widen the light receptive area by increasing the diameter of the objective lens to efficiently receive the fluorescence pumped or excited by the fluorescent substance, a light irradiating apparatus became large in size.

Further, in order to raise the detection accuracy of fluorescence by suppressing disturbance light (noise), it is necessary to approach the objective lens to the sample chip. But, there is a limitation in the approaching distance due to the physical properties of the objective lens, wherein the fluorescence could not be detected at high accuracy.

The present invention was developed to solve the abovementioned

problems and shortcomings in the prior art. It is therefore an object of the invention to provide a sample chip analyzing device and a method for analyzing the sample chip, which are capable of detecting, at high accuracy, fluorescence from marked fluorescent substances of a sample to be analyzed, which have been coupled to a sampling probe, without being influenced by pumping light of the fluorescent substance and disturbance light (noise), and efficiently analyzing the sample to be analyzed.

In addition, it is another object of the invention to provide a sample chip analyzing device and a method for analyzing the sample chip, which are capable of detecting fluorescence of different wavelengths at one time and efficiently analyzing the sample to be analyzed.

[Brief description of the drawings]

Fig.1 is a perspective view showing the entire sample chip analyzing device;

Fig.2 is a view showing the principle of the analyzing device;

Fig.3 is a view describing a waveguide of light on a glass sheet slide; and

Fig.4 is an enlarged view of part "A" in Fig.3.

[Embodiments of the invention]

Hereinafter, a description is given of embodiments of the invention with reference to the accompanying drawings.

In Fig.1 and Fig.3, a light shielding box 5 is installed at the side of a table 3 of a sample chip analyzing device 1. A light source 7 is

internally incorporated in the light shielding box 5. Also, an opening into which a waveguide plate 11 of a sample chip 9 described later is inserted in a close-adhered state is formed at the light shielding box 5, and the end portion of the waveguide plate 11 placed on a table 3 is inserted into the opening 13.

In the drawings, 12 denotes a packing adhered to the inner-circumferential surface of the opening 13, which prevents light from the light source 7 described later from leaking outside of the waveguide 11.

The light source 7 is determined by a wavelength that pumps a fluorescent substance marked on a sample to be analyzed, which is described later. For example, in the case where the marked fluorescent substance for the sample to be analyzed is single, the light source 7 may be a laser irradiating device that outputs light of a specified wavelength by which the corresponding fluorescent substance is pumped or in the case where a plurality of marked fluorescent substances are used, any one that outputs white light of respective pumping wavelengths may be acceptable.

The sample chip 9 set on the table 3 is such that a number of sampling probes 15 such as polynucleotide probes like DNA probes and RNA probes and protein peptide probes are fixed, at the waveguide plate 11 in which glass sheet slides or thin glass plates having an optical waveguide property, like dots at high density (several thousands to several ten thousands per square centimeter).

A pickup device 17 that photographs the surface of the sample

chip 9 is arranged above the sample chip 9 set on the table 3. As the corresponding pickup device 17, for example, a CCD camera is suitable, which can photograph the entire sample chip 9 as one frame or photograph the sample chip section by section with a plurality of frames.

Further, RGB filters is incorporated in the pickup device 17, and the corresponding RGB filters decomposes the pickup data color by color, and the color pickup data are stored in a buffer memory 19. And the central processing unit (CPU) 21 outputs the pickup data of the respective colors, which are stored in the buffer memory 19, and displays the entire image of the sample chip 9.

Next, a description is given of the method for analyzing a sample to be analyzed, by the abovementioned sample chip analyzing device.

The following description is based on a DNA analysis example. In Fig.3 and Fig.4, in order to hybridize a sample to be analyzed, on which a fluorescent substance is marked, with respect to the sampling probe 15 arrayed and fixed on the sample chip 9, a buffer solution containing a sample to be analyzed, which is extracted from cells and biological tissues, adjusted, and marked with a fluorescent substance, is adhered to the surface of sample chip 9, and is left in an appointed period of time as it is. At that time, where the sample to be analyzed and sampling probes 15 are complementary to each other, they are hybridized to become a two-chain structure. To the contrary, where they are not complementary to each other, they are not coupled to each other.

After the abovementioned appointed period of time elapses, the upper surface of the sample chip 9 is washed off by pure water or a buffer solution to remove the sampling probes 15 and non-coupled samples to be analyzed.

Next, after the sample chip 9 is set so that the end part thereof is inserted into the opening 13, light from the light source 7 is made incident from the end plane of the waveguide plate 11 into the interior thereof, whereby the light incident into the waveguide plate 11 is entirely reflected when the incident angle thereof exceeds the incident critical angle, and the light is guided to the other end part side of the sample chip.

At this time, an evanescent wave is generated on the surface and rear planes of the sample chip 9 by light that is entirely reflected in the interior of the waveguide plate 11, and a part thereof leaks out of the surface of the sample chip 9, wherein the electric field pumps the marked fluorescent substances of the sample to be analyzed, which is hybridized to the sampling probes 15 by the electric field, and the fluorescent substance fluoresces.

At this time, if the marked fluorescent substances of the sample to be analyzed, which is hybridized to respective sampling probes 15 of the sample chip 9, differ from each other, white light that is mixed by RGB may be used as the light source 7, and the respective fluorescent substances are caused to fluoresce by the respective colors. In this case, since the pumping energy of the respective fluorescent substances is low, it is necessary to increase the output of the light

source.

Using the pickup device 17, an operator picks up an image of the surface of the sample chip in which the marked fluorescent substances fluoresce, and displays it on a display device 23, wherein the fluoresced sampling probes are discriminated, the samples to be analyzed, which are hybridized to the sampling probes are specified and analyzed.

Since, in the abovementioned embodiment, the marked fluorescent substances of a sample to be analyzed, which are hybridized to the sampling probes 15 by an evanescent wave occurring when light advances in the interior of the waveguide plate 11 while being entirely reflected, are caused to fluoresce, there is no case where the reflected pumping light is received together with the fluorescence, in comparison with a prior art method in which light (pumping light) is irradiated from the outside to respective sampling probes and fluorescence from the fluorescent substances is received. Therefore, it is possible to shield influences of the pumping light and efficiently detect only the fluorescence, wherein it becomes possible to improve the analysis accuracy.

Also, in order to specify the samples to be analyzed, by discriminating the fluorescing sampling probes 15 from the pickup data on the surface of the sample chip 9, the time required for analysis can be remarkably shortened in comparison with the prior art method in which scanning is carried out by light.

What is claimed is:

1. A sample chip analyzing device, comprising:

a waveguide plate, which is able to entirely reflect incident light and guide the same, having a number of sampling probes on the surface thereof;

a light source for irradiating light from the end plane of said waveguide plate to the interior thereof; and

a pickup member for picking up an image of the surface of said waveguide plate on which said sampling probes are fixed;

wherein light is entirely reflected in the waveguide plate in which a sample to be analyzed, marked with fluorescent substances, is coupled to the sampling probes, the fluorescent substances are pumped by an evanescent wave occurring when guiding optical waves, and are caused to fluoresce, and a sample to be analyzed is able to be analyzed by an fluorescent image on the picked up surface of the waveguide plate.

2. A method for analyzing a sample chip, comprising the steps of:

pumping marked fluorescent substances of a sample to be analyzed, which are coupled to sampling probes by an evanescent wave occurring when making light from a light source incident into a waveguide plate, in which a number of sampling probes are fixed on the surface thereof, and guiding the same to the waveguide plate;

causing the marked fluorescent substances to fluoresce; and

analyzing said sample to be analyzed, on the basis of a fluorescent image on the picked up surface of said waveguide.

✓ 3. The sample chip analyzing device and method for analyzing a sample chip according to Claim 1[or 2] wherein said waveguide plate is



a glass substrate.

✓ 4. The sample chip analyzing device and method for analyzing the sample chip according to Claim 1 [or 2], wherein said waveguide plate has a pair of insulation reflection plates arranged opposite to each other with an appointed interval.

✓ 5. The sample chip analyzing device and method for analyzing the sample chip according to Claim 1 [or 2], wherein said light source outputs light of a wavelength that pumps the marked fluorescent substances.

✓ 6. The sample chip analyzing device and method for analyzing the sample chip according to Claim 1 [or 2], wherein said sampling probes and sample to be analyzed are made into any one of polynucleotide, peptide and protein.